REMARKS

Interview

Applicants would like to thank Examiner Kam for the phone conference held with Applicants' representative on May 18, 2006. During the phone interview, proposed amendments to the claims were discussed for overcoming the 35 U.S.C. § 112, first paragraph rejection. Applicants have considered and incorporated the amendments into the claims.

Status of the Claims

Claims 1, 2, 32-34, 37-45, 50-52, 57-61, and 63-68 are currently pending in the present application. Claims 3-31, 35, 36, 46-49, 53-56, and 62 have been canceled without prejudice or disclaimer of the subject matter claimed therein. Claims 1, 37, 50, 58, 59, and 64 have been amended.

Amendments to the Claims

The amendments to claims 1, 37, 50, 58, 59, and 64 do not introduce prohibited new matter.

Support for the amendments to claims 1 and 59 can be found throughout the specification. Representative support can be found on paragraph [0005], line 4 and paragraph [0015]. Applicants note that the exclusionary limitation added to these claims is merely to help define the claimed invention from the prior art. As such, this limitation is not prohibited new matter. *In re Johnson and Farnham*, 194 USPQ 187, 196 (CCPA 1977).

Claim 37 has been amended to correct a typographical error and to remove "amyloid β-protein" from the claim in view of the amendment to claim 1.

Claims 50 and 58 have been amended to be consistent with the rest of the claims.

Claim 64 has been amended to change its dependency to claim 59, since claim 3 has been canceled.

Objection to Claim 37

Claim 37 has been objected to for reciting improper Greek characters. Claim 37 has been amended to correct the inadvertent typographical mistake.

Rejection Under 35 U.S.C. 112, first paragraph

Claims 1-3, 32-34, 37-55, and 57-68 are rejected under 35 U.S.C. § 112, first paragraph, because the specification does not enable a method of removing amyloid deposits from a subject comprising administering to the subject any amyloid fibril, a pharmaceutical composition formulated for removing amyloid deposits from a subject comprising any amyloid fibrils, or a vaccine composition comprising any amyloid fibrils.

Without acquiescing to the propriety of the rejection, claims 3, 46-49, 53-55, and 62, directed to vaccine compositions, have been canceled without prejudice or disclaimer of the subject matter claimed therein. Claims 1, 2, 32-34, 37-52, 57-61, and 63-68 are directed to a method of removing amyloid deposits from a subject comprising administering to the subject amyloid fibrils in an effective amount to generate an immune response, wherein the immune response promotes the removal of amyloid deposits from the subject and wherein the amyloid fibrils do not comprise amyloid β-protein.

Applicants respectfully point out that the initial burden is on the Examiner to provide a reasonable explanation as to why the scope of protection provided by the claim is not adequately enabled by the disclosure. *In re Wright*, 999 F.2d 1557, 1562, 27 USPQ2d 1510, 1513 (Fed. Cir. 1993). Moreover, the court in *In re Marzocchi* stated that it is incumbent upon the Patent Office to explain why it doubts the truth or accuracy of any statement in a supporting disclosure and to back up assertions of its own with acceptable evidence or reasoning which is inconsistent with the contested statement. *In re Marzocchi*, 439 F.2d 220, 224, 169 USPQ 367, 370 (CCPA 1971). The Office Action has not provided any reason to doubt the enablement of the claimed invention. Moreover, the Office Action has not provided a reasonable explanation or evidence establishing the nonenablement of the claims. In the absence of evidence to the contrary, the specification fully enables the claims directed to a method of removing amyloid deposits from a

subject comprising administering to the subject amyloid fibrils in an effective amount to generate an immune response, wherein the amyloid fibrils do not comprise amyloid β-protein.

The Office Action alleges that the specification only provides cursory conclusions without data supporting the findings. Applicants respectfully point out that Example D on page 35 discloses data supporting the removal of amyloid deposits from mice. First, the mice were immunized with synthetic fibrils and were shown to have anti-fibril antibodies. Subsequently, the mice were subcutaneously injected with human AL amyloid extract to produce a huge AL amyloidoma. Since the mice were immunized with synthetic fibrils prior to injection with the human AL amyloid extract, the amyloidoma disappeared in 5 days as compared to more than 15 days in non-vaccinated control mice. Thus the specification provides data to support the findings.

Furthermore, the attached reference of Schell *et al.* (Prevention of AA Amyloidosis by Active Immunotherapy. Amyloid and Amyloidosis 2001. Miklos Bely and Agnes Apathy Eds. Agnes Apathy Publishing, Budapest, pp.234-235) provides evidence that amyloid fibrils heterologous to the amyloid fibrils in the amyloid deposits of a subject can remove the amyloid deposits from the subject. Specifically, Schell *et al.* show that amyloid fibrils obtained from light chain immunoglobulin are capable of removing amyloid deposits in AA-amyloidotic mice. The amyloid fibrils administered to the subject are heterologous to the amyloid fibrils in the amyloid deposits of a subject in terms of the precursor proteins from which they are derived. As discussed in detail in the previous response, dated February 22, 2005, amyloid fibrils are structurally homologous molecules.

The Office Action also alleges that the specification does not enable the full scope of the claims and cites the following Wands factor for determining whether undue experimentation is required.

1. The breadth of the claims:

The Office Action alleges that the claims are broad in scope and encompass unspecified variants of amyloid fibrils contained in a pharmaceutical composition used in the removal of

amyloid deposits from a subject, which are not adequately described or demonstrated in the specification. Applicants respectfully point out that the claimed method of removing amyloid deposits from a subject can be accomplished with any amyloid fibrils because amyloid fibrils, irrespective of the precursor protein that they are made from, are structurally homologous molecules and thereby elicit a generic anti-fibril immune response. In the response dated February 22, 2005, Applicants discussed in detail the data from different research groups showing amyloid fibrils are structurally homologous molecules. Moreover, in the same response, Applicants provided evidence to demonstrate that the AA-amyloidosis murine model used in the present application is an accepted animal model for studying amyloidosis in human and that the AA-amyloidosis mice is representative of the disease in humans.

Accordingly, since amyloid fibrils are structurally homologous and the murine model used by Applicants is an accepted animal model for studying amyloidosis in human and since there is no reason to doubt the enablement of the disclosed vaccines, Applicants have enabled the breadth of the claims.

2. The absence and presence of working examples:

The Office Action alleges that the specification has not demonstrated that pharmaceutical compositions comprising amyloid fibrils are capable of removing amyloid deposits in mice with AA-amyloidosis. As discussed previously, Example D shows that synthetic amyloid fibrils administered as a vaccine to mice in an art accepted murine model resulted in the removal of the amyloidoma. The Example also teaches the removal of AA amyloid from mice induced to develop systemic AA amyloidosis. Also as discussed above, these murine models are well known and accepted as animal models for studying human amyloidosis. Moreover, the specification in paragraph 0018 discusses in detail animal models for amyloidosis. Additionally, it is well known that amyloid fibrils are structurally homologous (see specification paragraphs 0005 and 0132) molecules that can be used to induce a cross-reactive immune response.

Further, the attached reference of Schell *et al.* provides evidence that amyloid fibrils heterologous to the amyloid fibrils in the amyloid deposits of a subject can remove the amyloid

deposits from the subject. Specifically, Schell *et al.* show that amyloid fibrils obtained from light chain immunoglobulin are capable of removing amyloid deposits in AA-amyloidotic mice.

Thus, there are sufficient examples to enable the scope of the claims.

3. The state of the prior art and relative skills of those in the art; and 4. The predictability and unpredictability of the art:

The Office Action alleges that the art does not disclose the use of amyloid fibril components which are heterologous to amyloid fibrils to treat patients and that the general knowledge and skill of those in the art are not sufficient to enable the scope of the claimed invention. Applicants respectfully submit that given the general knowledge of the murine mouse models for amyloidosis and the structural properties and similarities of amyloid fibrils, and given the guidance in the specification for administering amyloid fibrils to subjects (pages 22-25) and for methods of treating amyloidosis (pages 6 and 7), there is sufficient guidance to enable the scope of the claims.

The Office Action alleges that the specification has not shown that pharmaceutical composition comprising amyloid fibrils are capable of removing amyloid deposits in mice with AA-amyloidosis. Applicants respectfully point out that as taught in the specification, a specific amyloid fibril need not be administered to a subject to remove a specific type of deposit, because as discussed above, amyloid fibrils, irrespective of the precursor proteins from which they are made, are structurally related molecules. As shown in Example D, Applicants unexpectedly discovered that a synthetic amyloid fibril comprising immunoglobulin light chain variable region domain was capable of inducing an immune response sufficient to remove an *in vivo* murine amyloidoma generated by injecting human AL amyloid extract into a mouse. As discussed above and taught in the specification, since amyloid fibrils are structurally homologous molecules, the immune response induced by the synthetic amyloid fibril was sufficient to remove the murine amyloidoma generated from human AL amyloid extract by virtue of its reactivity with the generic, shared amyloid fibril epitopes. Moreover, the claimed invention can be performed with

any amyloid fibril whether synthetic or naturally occurring as a consequence of the structural identity of amyloid fibrils.

Further as discussed above, the reference of Schell *et al.* confirms that amyloid fibrils heterologous to the amyloid fibrils in the amyloid deposits of a subject can remove the amyloid deposits from the subject. Specifically, Schell *et al.* show that amyloid fibrils obtained from light chain immunoglobulin are capable of removing amyloid deposits in AA-amyloidotic mice.

Given the guidance provided by the specification and given the state of the prior art disclosing amyloidosis and the structural properties of amyloid fibrils, the present invention is neither unpredictable nor lacks enablement for the scope of the claims.

5. The amount of direction or guidance presented and the quantity of experimentation necessary and 6. The nature of the invention:

The Office Action alleges that the specification has not demonstrated that pharmaceutical composition comprising amyloid fibrils are capable of removing amyloid deposits from mice with AA-amyloidosis and has not demonstrated the effect of amyloid fibrils heterologous to amyloid fibrils in the amyloid deposits of a subject.

As discussed above, the reference of Schell *et al.* provides evidence that amyloid fibrils heterologous to the amyloid fibrils in the amyloid deposits of a subject can remove the amyloid deposits from the subject. Specifically, Schell *et al.* show that amyloid fibrils obtained from light chain immunoglobulin are capable of removing amyloid deposits in AA-amyloidotic mice.

Also, as discussed above, the specification teaches that amyloid fibrils are structurally homologous molecules and that immunization of mice with an amyloid fibril generates an immune response that promotes the removal of amyloid deposits, including deposits comprising fibrils whose precursor molecules are distinct from those of the amyloid fibrils used to immunize the mice.

Moreover, as shown in Example D, a synthetic amyloid fibril comprising immunoglobulin light chain variable domain was capable of inducing an immune response

sufficient to remove an *in vivo* murine amyloidoma generated by injecting human AL amyloid extract into a mouse.

In view of the guidance provided by the specification, including Example D, and the state of the art, there is adequate direction and guidance to enable the scope of the invention.

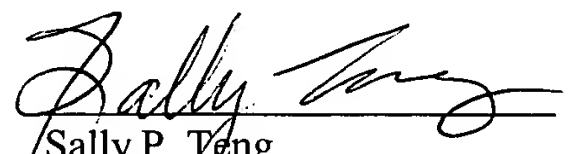
Applicants respectfully point out that in response to the Office Action, dated February 22, 2005, the rejection under 35 U.S.C. 112 § first paragraph was withdrawn with respect to claims 1, 2, 32-45, 50, 52, 56, and 57 in the subsequent Office Action, dated June 27, 2005. Accordingly, Applicants request withdrawal of the present rejection.

Conclusion

In view of the foregoing claim amendments and accompanying remarks, Applicants respectfully request reconsideration and timely allowance of the pending claims. Should the Examiner feel that there are any issues outstanding after consideration of this response, the Examiner is invited to contact Applicants' undersigned representative to expedite prosecution.

If there are any additional fees due in connection with the filing of this response, please charge the fees to our Deposit Account No. 50-0310. If a fee is required for an extension of time under 37 C.F.R. § 1.136 not accounted for above, such an extension is requested and the fee should also be charged to our Deposit Account.

Respectfully submitted,
Morgan, Lewis & Bockius LLP

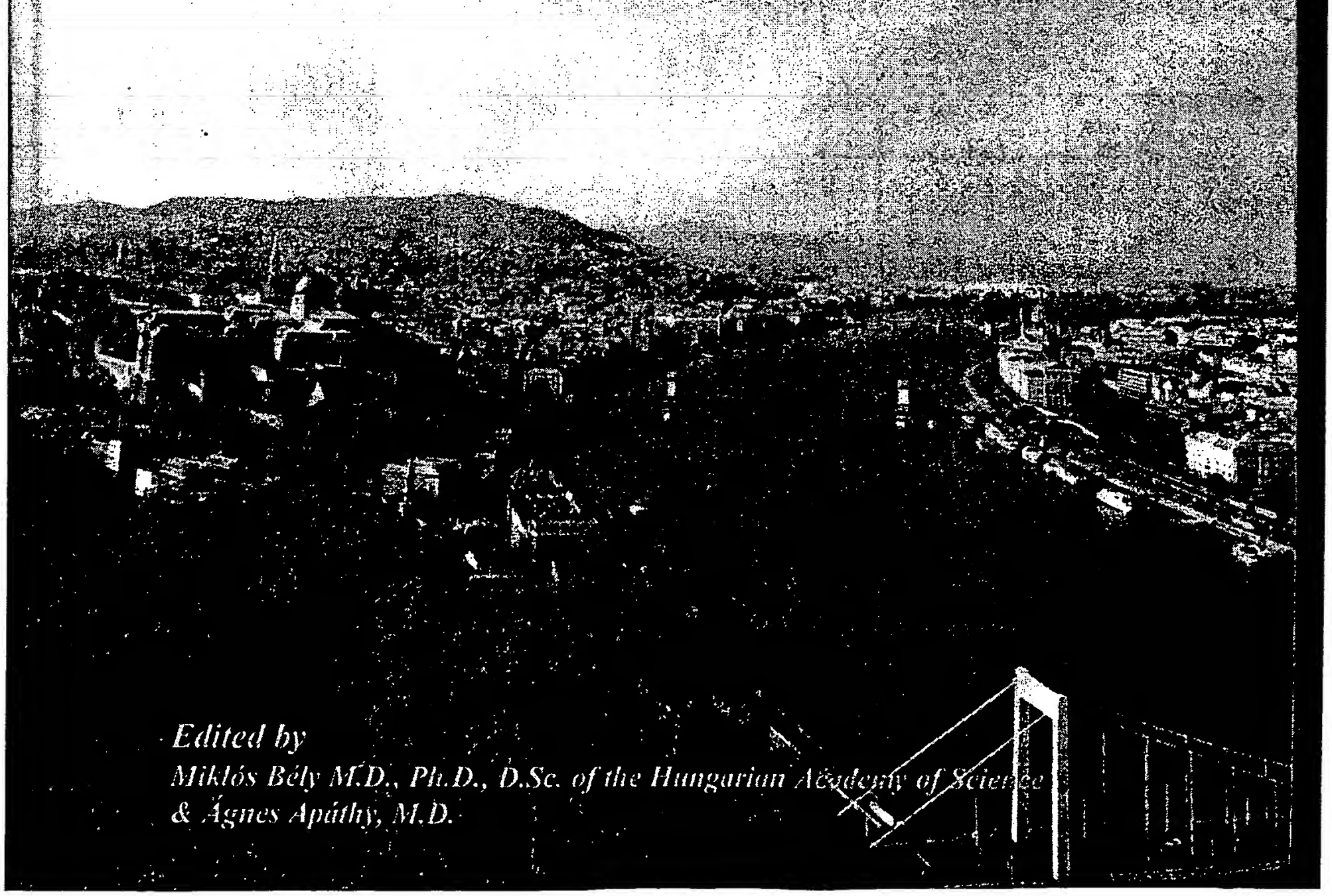


Sally P. Teng
Registration No. 45,397

Dated: **June 19, 2006**
Morgan, Lewis & Bockius LLP
Customer No. **09629**
1111 Pennsylvania Avenue, N.W.
Washington, D.C. 20004
202-739-3000

Amyloid and Amyloidosis

The Proceedings of the IXth International Symposium on Amyloidosis



Edited by
Miklós Bély M.D., Ph.D., D.Sc. of the Hungarian Academy of Science
& Ágnes Apáthy, M.D.

PREVENTION OF AA AMYLOIDOSIS BY ACTIVE IMMUNOTHERAPY

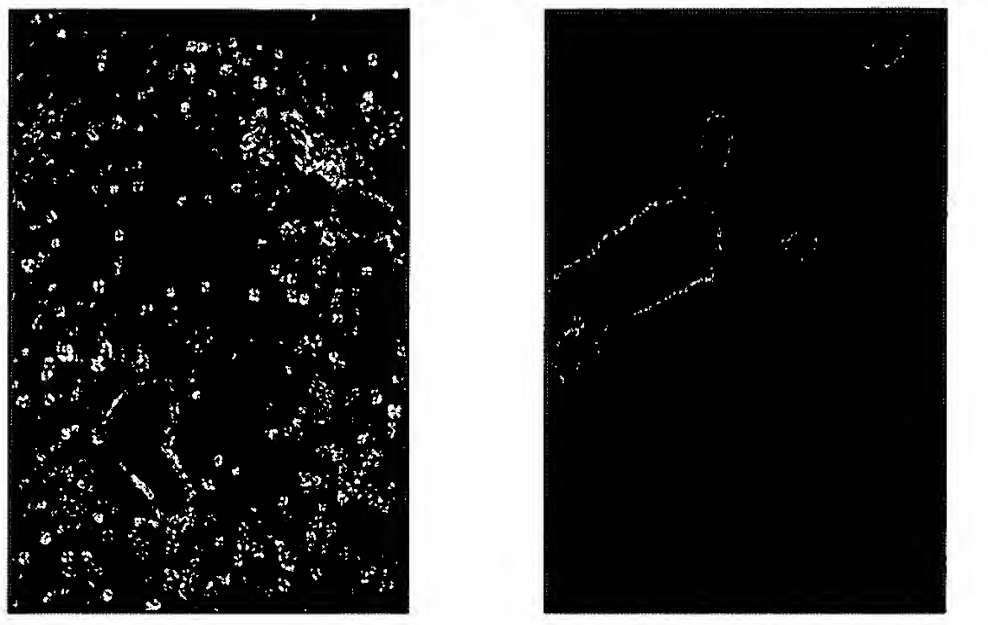
Maria Schell, Jonathan Wall, Sallie Macy, Craig Wooliver, Dennis Wolfenbarger, Robert Donnell, Deborah Weiss and Alan Solomon
Address: Human Immunology & Cancer Program, Graduate School of Medicine, University of Tennessee, 1924 Alcoa Hwy, Knoxville TN 37920, USA

Introduction: A novel murine model of AA-amyloidosis has been used in our laboratory that permits the study of pathological effects of continuous expression of cytokines. Transgenic mice (C57BL/6J strain) were generated that express the human IL-6 gene under the control of the mouse metallothionein-1 (MT-1) promoter. The hIL-6 mice have increased levels of acute phase proteins, B-cell hyperplasia in lymph-nodes and spleen, a polyclonal increase in serum IgG with renal tubular casts[1]. We found that these animals develop systemic amyloidosis at 3 months of age and the disease is progressive up to 8 or 9 months when the animals become visibly ill and either succumb or have to be euthanized[2]. The fibrillar deposits occur primarily in the spleen, liver and kidneys and contain AA-related amyloid as evidenced immunohistochemically and by amino acid sequence and mass spectroscopic analyses of the extracted protein[2]. We have now shown that amyloid formation in IL-6 transgenic mice can be accelerated by administration of AA amyloid enhancing factor (AEF) derived from human or murine amyloid-laden tissue[3,4]. A single *iv* injection of 10 μg of AEF in the tail vein of 6 wk-old animals resulted in readily detectable amyloid deposits in the liver, spleen and kidney within 2 wks. Over an additional 8 to 9 wks the mice developed massive amyloid deposits and due to morbidity were sacrificed. This experimental model, designated TRIAD (transgenic rapidly inducible amyloid deposition), has been employed to evaluate a novel immune-based therapy for AA amyloidosis.

Materials and Methods: Human λ light chain protein ($rV_\lambda 6$) was produced in our laboratory using a bacterial pET expression system (Novagen, Inc., Madison, WI). Synthetic amyloid fibrils were prepared by shaking a solution of the recombinant light chain protein (1 mg/ml in PBS) at 37 °C , 225 rpm for 36 hours. Amyloid fibrils were collected by centrifugation at 17000x g and similarly washed ×3 in distilled water. They were suspended in PBS at a concentration of 2 mg/ml, sonicated, and mixed with Imjet®Alum (Pierce, Rockford, IL) at a ratio 1:1. This preparation was used as the immunogen.

Groups of 8 wk-old hIL-6 transgenic mice were injected i.p. with 100 μg of immunogen plus alum, while control animals received PBS and alum alone. The injections were repeated weekly ×3 followed by 2 booster injections 2 wks apart, for a total of 5 injections over a 7 wk period. Serum anti-$rV_\lambda 6$fibril reactivity was tested by ELISA using sera from immunized mice as the primary antibody. One week after the final injection, all mice received a single *iv* injection of 10 μg of AEF and the animals were sacrificed 3, 6, or 10 weeks later.

Sera from the immunized mice were tested immunohistochemically for anti-AA amyloid reactivity using sections of amyloid-laden liver and spleen cut from formalin-fixed, paraffin-embedded blocks of tissue obtained from the TRIAD mice. For these studies we used the biotin-streptavidin (B-SA) technique (BioGenex, San Ramón, CA) and "The Mouse-on-Mouse" Iso-IHC kit (InnoGenex San Ramón, CA). Histological analyses were performed using; Congo red, naphthol AS-D, naphthyl acetate esterase, and periodic acid Schiff's stains (Sigma Diagnostics, St. Louis, MO) according the manufacturer's specifications. Image analysis and spectral segmentation techniques were used to quantitate the amyloid burden index (ABI) in Congo red-stained tissue sections of spleen and liver.



Control (3.8 % amyloid burden) Immunized (0.24 % amyloid burden)

Figure 1. Quantitation of hepatic amyloid in control and immunized MT/hIL-6 mice

Results: By ELISA, the sera of immunized mice contained low titers of anti-rV_λ6 fibril antibodies that reacted in immunohistochemical studies with AL, as well as with AA amyloid. No such activity was detected in the sera from non-immunized mice. Of the control animals, 50% of the mice died within 3 wks and 100% at 10 wks post-AEF injection due to extensive systemic AA deposition; in contrast, all of the immunized mice were alive at 10 wks. Quantitation of the amyloid burden in liver

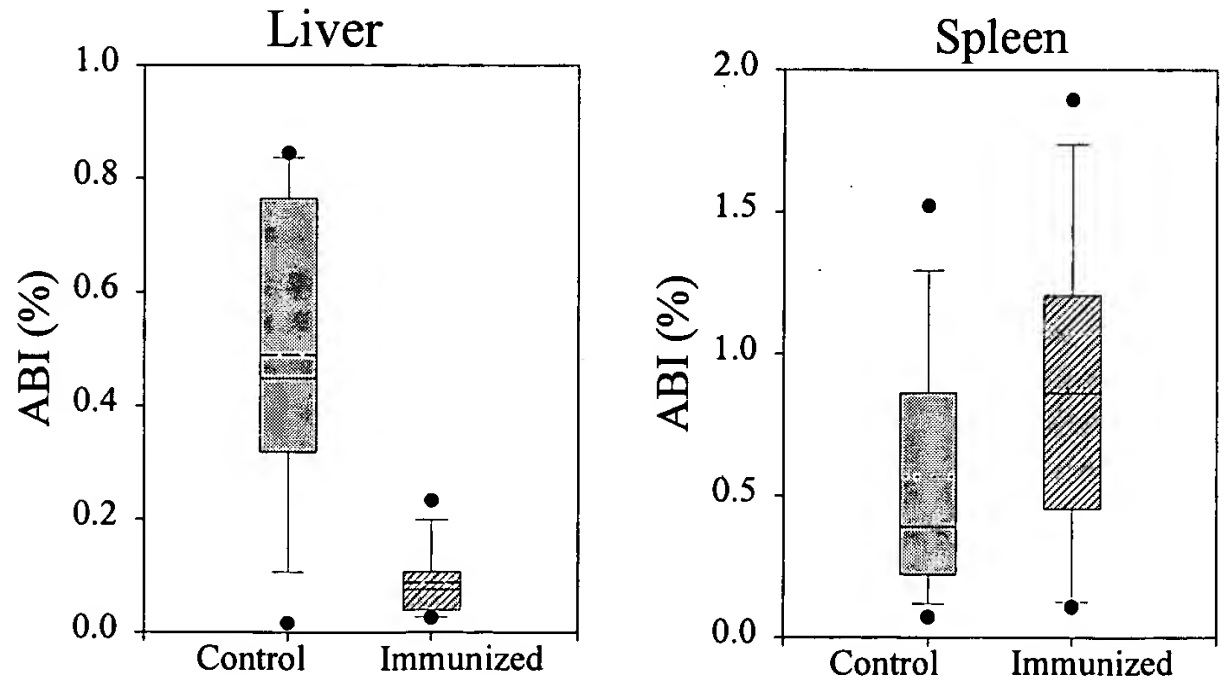


Figure 2. Comparison of hepatic and splenic amyloid burden indices (ABI) for immunized and control mice. The dashed and solid lines are the mean and median, respectively. The boxes represent the 25th and 75th percentile and the error bars the 10th and 90th percentiles.

sections from these animals revealed an 80% reduction with respect to the controls (Figs 1 and 2). There was no significant difference in the quantity of splenic amyloid in the treated vs. the control group (Fig 2).

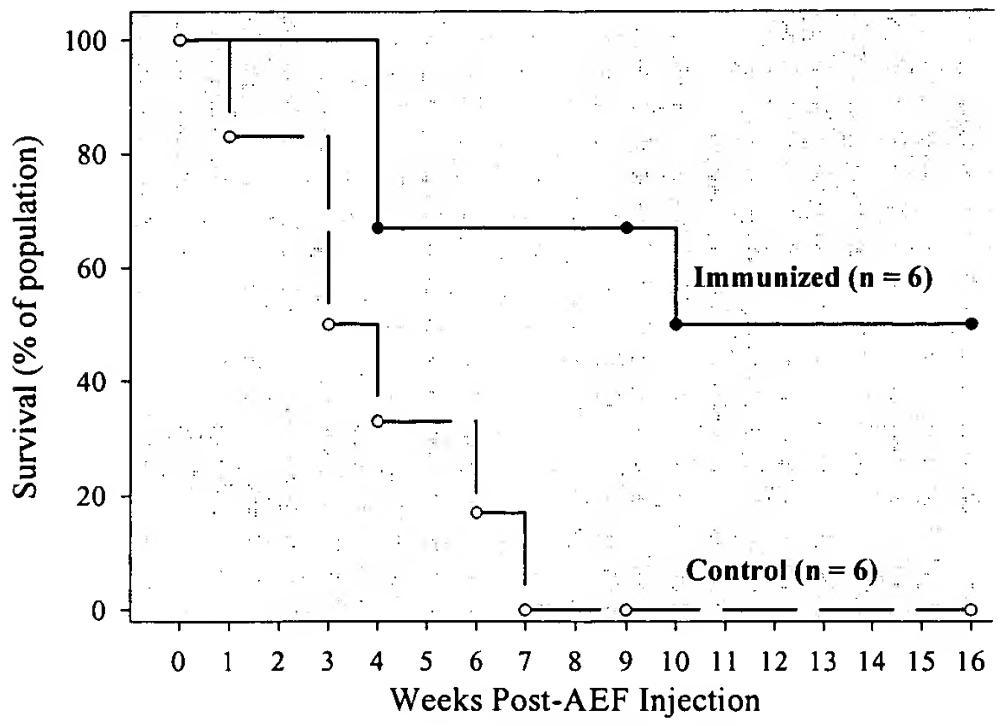


Figure 3. Survival of immunized and control AA-amyloidotic mice

Examination of liver tissue from immunized mice stained with a dual esterase reagent and PAS revealed intrinsic hepatocyte esterase function and the presence of glycogen granules, respectively, both of which were absent in mice that had extreme hepatic amyloid deposits. Additionally, we found that immunization prolonged long-term survival: 50% of the immunized mice were alive at 16 wks post-AEF injection as compared to controls that were all deceased by wk 7 (Fig. 2).

Conclusion: Our studies have demonstrated that AA-related deposition was markedly reduced in the liver of mice immunized with light chain synthetic fibrils. Based on these findings, we suggest that this heterogeneous vaccine-based strategy may be used to prevent AA and increase long-term survival of those with the disease, as well as other forms of amyloidosis.

References:

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4. Shtrasburg S, Livneh A, Gal R and Pras M (1996) Extremely active murine amyloid enhancing factor. *Clin Exp Rheumatol*, **14**, 37-42